

09/402093

514 Rec'd REC'D CT/PTO 29 SEP 1999

SEQUENCE LISTING

<110> Suntory Limited  
  
<120> Process for producing peptides using a helper peptide  
  
<130> F962  
  
<150> JP 10-032272  
<151> 1998-01-30  
  
<160> 24  
  
<210> 1  
<211> 4  
<212> PRT  
<213> Artificial Sequence  
  
<220>  
<223> Amino acid sequence adjacent to a site cleaved by enterokinase  
  
<400> 1  
Asp Asp Asp Lys  
1  
  
<210> 2  
<211> 4  
<212> PRT  
<213> Artificial Sequence  
  
<220>  
<223> Amino acid sequence adjacent to a site cleaved by blood coagulation Factor Xa

<400> 2  
Ile Glu Gly Arg  
1

<210> 3  
<211> 7  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Amino acid sequence containing a site cleaved by renin

<400> 3  
Pro Phe His Leu Leu Val Tyr  
1 5

<210> 4  
<211> 6  
<212> PRT  
<213> Artificial Sequence

<220>  
<223>

<400> 4  
Val Asp Asp Asp Asp Lys  
1 5

<210> 5  
<211> 6  
<212> PRT  
<213> Artificial Sequence

<220>

<223> Amino acid sequence of helper peptide

<400> 5

Gly Cys His His His His

1 5

<210> 6

<211> 10

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence containing a chemically cleaved site

<400> 6

Pro Gly Gly Arg Pro Ser Arg His Lys Arg

1 5 10

<210> 7

<211> 10

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence of helper peptide

<400> 7

His Arg His Lys Arg Ser His His His His

1 5 10

<210> 8

<211> 5

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence containing a site cleaved by Kex2 protease

<400> 8

Ser Asp His Lys Arg

1 5

<210> 9

<211> 23

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence of containing a position cleaved by OmpT

<400> 9

Gln Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His

1 5 10 15

Arg Trp Gly Arg Ser Gly Ser

20

<210> 10

<211> 20

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence containing a position cleaved by OmpT

<400> 10

Gln Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His  
1 5 10 15

Gly Ser Gly Ser  
20

<210> 11  
<211> 69  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Nucleotide sequence coding for an amino acid sequence containing a site cleaved by OmpT

<400> 11  
cag atg cat ggt tat gac gcg gag ctc cgg ctg tat cgc cgt cat cac 48  
Gln Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His  
1 5 10 15  
cgg tgg ggt cgt tcc gga tcc 69  
Arg Trp Gly Arg Ser Gly Ser  
20

<210> 12  
<211> 23  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Amino acid sequence containing a site cleaved by OmpT

<400> 12  
Gln Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His  
1 5 10 15  
Arg Trp Gly Arg Ser Gly Ser  
20

[REDACTED]

<210> 13  
<211> 47  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Nucleotide sequence coding for an amino acid sequence containing a site cleaved by OmpT

<400> 13  
tggttatgac gcggagctcc gcctgtatcg ccgtcatcac gttccg 47

<210> 14  
<211> 55  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Nucleotide sequence coding for an amino acid sequence containing a site cleaved by OmpT

<400> 14  
gatccggaac cgtgatgacg gcgatacagg cggagctccg cgtcataacc atgca 55

<210> 15  
<211> 24  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Primer

<400> 15  
gactcagatc ttcctgaggc cgat 24

<210> 16  
<211> 36  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Primer

<400> 16  
aaaggtacct tccgcattgcc gcggatgtcg agaagg 36

<210> 17  
<211> 20  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Primer

<400> 17  
aggccaggaa ccgtaaaaag 20

<210> 18  
<211> 29  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Primer

<400> 18  
aaaatgcatac gcatcgtaac cgtgcatact 29

<210> 19

<211> 627  
<212> DNA  
<213> Artificial Sequence

<220>  
<223> Nucleotide sequence coding for a fusion protein comprising GLP-1, helper peptide and β-galactosidase protective peptide

<400> 19

cccaggctt acactttatg cttccggctc gtatgttgtg tgaaattgtg agcgataac	60
aatttcacac aggaaacagc t atg acc atg att acg gat tca ctg gcc gtc	111
Met Thr Met Ile Thr Asp Ser Leu Ala Val	
1 5 10	
gtt tta caa cgt aaa gac tgg gat aac cct ggc gtt acc caa ctt aat	159
Val Leu Gln Arg Lys Asp Trp Asp Asn Pro Gly Val Thr Gln Leu Asn	
15 20 25	
cgc ctt gca gca cat ccc cct ttc gcc agc tgg cgt aat agc gac gac	207
Arg Leu Ala Ala His Pro Pro Phe Ala Ser Trp Arg Asn Ser Asp Asp	
30 35 40	
gcc cgc acc gat cgc cct tcc caa cag ttg cgc agc ctg aat ggc gaa	255
Ala Arg Thr Asp Arg Pro Ser Gln Gln Leu Arg Ser Leu Asn Gly Glu	
45 50 55	
tgg cgc ttt gcc tgg ttt ccg gca cca gaa gcg gtg ccg gca agc ttg	303
Trp Arg Phe Ala Trp Phe Pro Ala Pro Glu Ala Val Pro Ala Ser Leu	
60 65 70	
ctg gag tca gat ctt cct gag gcc gat act gtc gtc gtc ccc tca aac	351
Leu Glu Ser Asp Leu Pro Glu Ala Asp Thr Val Val Val Pro Ser Asn	
75 80 85 90	
tgg cag atg cac ggt tac gat gcg atg cat ggt tat gac gcg gag ctc	399
Trp Gln Met His Gly Tyr Asp Ala Met His Gly Tyr Asp Ala Glu Leu	
95 100 105	
cgc ctg tat cgc cgt cat cac ggt tcc gga tcc cct tct cga cat ccg	447
Arg Leu Tyr Arg Arg His His Gly Ser Gly Ser Pro Ser Arg His Pro	
110 115 120	

cgg cat gcg gaa ggt acc ttt acc agc gat gtg agc tcg tat ctg gaa 495  
Arg His Ala Glu Gly Thr Phe Thr Ser Asp Val Ser Ser Tyr Leu Glu  
125 130 135  
ggt cag gcg gca aaa gaa ttc atc gcg tgg ctg gtg aaa ggc cgt ggt 543  
Gly Gln Ala Ala Lys Glu Phe Ile Ala Trp Leu Val Lys Gly Arg Gly  
140 145 150  
taagtcgaca gccccctaa tgagcgggct ttttttctc ggaattaatt ctcatgttg 603  
acagcttatac atcgataaagc ttta 627

<210> 20  
<211> 154  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Amino acid sequence of a fusion protein comprising  
GLP-1, helper peptide and  $\beta$ -galactosidase  
protective peptide

<400> 20  
Met Thr Met Ile Thr Asp Ser Leu Ala Val Val Leu Gln Arg Lys Asp  
1 5 10 15  
Trp Asp Asn Pro Gly Val Thr Gln Leu Asn Arg Leu Ala Ala His Pro  
20 25 30  
Pro Phe Ala Ser Trp Arg Asn Ser Asp Asp Ala Arg Thr Asp Arg Pro  
35 40 45  
Ser Gln Gln Leu Arg Ser Leu Asn Gly Glu Trp Arg Phe Ala Trp Phe  
50 55 60  
Pro Ala Pro Glu Ala Val Pro Ala Ser Leu Leu Glu Ser Asp Leu Pro  
65 70 75 80  
Glu Ala Asp Thr Val Val Val Pro Ser Asn Trp Gln Met His Gly Tyr  
85 90 95  
Asp Ala Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His  
100 105 110  
His Gly Ser Gly Ser Pro Ser Arg His Pro Arg His Ala Glu Gly Thr  
115 120 125

Phe Thr Ser Asp Val Ser Ser Tyr Leu Glu Gly Gln Ala Ala Lys Glu  
130 135 140

Phe Ile Ala Trp Leu Val Lys Gly Arg Gly  
145 150

<210> 21  
<211> 187  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Amino acid sequence of a fusion protein comprising  
GLP-1, helper peptide and  $\beta$ -galactosidase  
protective peptide

<400> 21  
Met Thr Met Ile Thr Asp Ser Leu Ala Val Val Leu Gln Arg Lys Asp  
1 5 10 15  
Trp Asp Asn Pro Gly Val Thr Gln Leu Asn Arg Leu Ala Ala His Pro  
20 25 30  
Pro Phe Ala Ser Trp Arg Asn Ser Asp Asp Ala Arg Thr Asp Arg Pro  
35 40 45  
Ser Gln Gln Leu Arg Ser Leu Asn Gly Glu Trp Arg Phe Ala Trp Phe  
50 55 60  
Pro Ala Pro Glu Ala Val Pro Ala Ser Leu Leu Glu Ser Asp Leu Pro  
65 70 75 80  
Glu Ala Asp Thr Val Val Val Pro Ser Asn Trp Gln Met His Gly Tyr  
85 90 95  
Asp Ala Pro Ile Tyr Thr Asn Val Thr Tyr Pro Ile Thr Val Asn Pro  
100 105 110  
Pro Phe Val Pro Thr Glu Pro His His His His His Gly Arg Gln  
115 120 125  
Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His Arg  
130 135 140  
Trp Gly Arg Ser Gly Ser Pro Ser Arg His Lys Arg His Ala Glu Gly  
145 150 155 160

Thr Phe Thr Ser Asp Val Ser Ser Tyr Leu Glu Gly Gln Ala Ala Lys  
165 170 175

Glu Phe Ile Ala Trp Leu Val Lys Gly Arg Gly  
180 185

<210> 22

<211> 184

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence of a fusion protein comprising  
GLP-1, helper peptide and  $\beta$ -galactosidase  
protective peptide

<400> 22

Met Thr Met Ile Thr Asp Ser Leu Ala Val Val Leu Gln Arg Lys Asp  
1 5 10 15

Trp Asp Asn Pro Gly Val Thr Gln Leu Asn Arg Leu Ala Ala His Pro  
20 25 30

Pro Phe Ala Ser Trp Arg Asn Ser Asp Asp Ala Arg Thr Asp Arg Pro  
35 40 45

Ser Gln Gln Leu Arg Ser Leu Asn Gly Glu Trp Arg Phe Ala Trp Phe  
50 55 60

Pro Ala Pro Glu Ala Val Pro Ala Ser Leu Leu Glu Ser Asp Leu Pro  
65 70 75 80

Glu Ala Asp Thr Val Val Val Pro Ser Asn Trp Gln Met His Gly Tyr  
85 90 95

Asp Ala Pro Ile Tyr Thr Asn Val Thr Tyr Pro Ile Thr Val Asn Pro  
100 105 110

Pro Phe Val Pro Thr Glu Pro His His His His His Gly Arg Gln  
115 120 125

Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His Gly  
130 135 140

Ser Gly Ser Pro Ser Arg His Lys Arg His Ala Glu Gly Thr Phe Thr  
145 150 155 160

Ser Asp Val Ser Ser Tyr Leu Glu Gly Gln Ala Ala Lys Glu Phe Ile  
165 170 175

Ala Trp Leu Val Lys Gly Arg Gly  
180

<210> 23  
<211> 184  
<212> PRT  
<213> Artificial Sequence

<220>  
<223> Amino acid sequence of a fusion protein comprising  
GLP-1, helper peptide and  $\beta$ -galactosidase  
protective peptide

<400> 23

Met Thr Met Ile Thr Asp Ser Leu Ala Val Val Leu Gln Arg Lys Asp  
1 5 10 15

Trp Asp Asn Pro Gly Val Thr Gln Leu Asn Arg Leu Ala Ala His Pro  
20 25 30

Pro Phe Ala Ser Trp Arg Asn Ser Asp Asp Ala Arg Thr Asp Arg Pro  
35 40 45

Ser Gln Gln Leu Arg Ser Leu Asn Gly Glu Trp Arg Phe Ala Trp Phe  
50 55 60

Pro Ala Pro Glu Ala Val Pro Ala Ser Leu Leu Glu Ser Asp Leu Pro  
65 70 75 80

Glu Ala Asp Thr Val Val Val Pro Ser Asn Trp Gln Met His Gly Tyr  
85 90 95

Asp Ala Pro Ile Tyr Thr Asn Val Thr Tyr Pro Ile Thr Val Asn Pro  
100 105 110

Pro Phe Val Pro Thr Glu Pro His His His His His Gly Arg Gln  
115 120 125

Met His Gly Tyr Asp Ala Glu Leu Arg Leu Tyr Arg Arg His His Gly  
130 135 140

Ser Gly Ser Pro Ser Arg His Pro Arg His Ala Glu Gly Thr Phe Thr  
145 150 155 160

Ser Asp Val Ser Ser Tyr Leu Glu Gly Gln Ala Ala Lys Glu Phe Ile  
165 170 175

Ala Trp Leu Val Lys Gly Arg Gly  
180

<210> 24

<211> 5

<212> PRT

<213> Artificial Sequence

<220>

<223> Amino acid sequence containing a site cleaved by  
Kex2 Protease

<400> 24

Ser Cys His Lys Arg

1 5